

## SHORT COMMUNICATION

# Short-term residual N unaffected by forbs in grass-clover mixtures

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## Abstract

We determined the effect on residual nitrogen (N) of including forbs (chicory, ribwort plantain and caraway) in perennial ryegrass-red clover mixtures. Although soil N inputs during the grassland phase differed markedly between mixtures, in a pot experiment we found no differences in the potentially mineralizable N of the soil or in the dry matter production and N content of the spring barley test crop. The fertilizer value of the grassland mixtures corresponded to 10 g N/m<sup>2</sup>, irrespective of forb inclusion. Thus, the inclusion of nonlegume forbs did not negatively affect short-term residual N fertility of legume-based grasslands.

**Keywords:** Grassland, residual effect, chicory, caraway, ribwort plantain

## Introduction

Recently, nonlegume forbs have been included in grass-clover mixtures to improve mineral nutrition for grazing animals from the forage (Pirhofer-Walzl *et al.*, 2011), plant diversity and herbage production (Cong *et al.*, 2017). In grasslands, legume biomass is an indicator of N released to the subsequent crop (Vrignon-Brenas *et al.*, 2016); hence, inclusion of nonlegume forbs could affect the residual N fertility due to their replacement of N-rich legumes in the sward. We investigated how inclusion of forbs would influence the residual N effect of grass-clover mixtures.

## Materials and methods

The grasslands, established in spring 2013 in a long-term organic dairy crop rotation on loamy sand (Eriksen *et al.*, 2015), were composed of red clover (*Trifolium pratense* L., variety Rajah), perennial ryegrass (*Lolium perenne* L., variety Stefani), chicory (*Cichorium intybus* L., variety Spadona), ribwort plantain (*Plantago lanceolata* L., wild type) and caraway (*Carum carvi* L., variety Volhouden) in different seeding mixtures (Table 1) and managed as described in Cong *et al.* (2017). The resultant swards varied in red clover

proportion, input of N from N<sub>2</sub>-fixation (246–617 kg N/ha), and N balance (–54 to 137 kg N/ha) depending on species composition in the seed mixtures (Table 1).

In spring 2015, 50 kg soil was collected from the plough layer (0–20 cm) of each treatment. Plant residues were separated from the soil, cut into 0.5 cm pieces, and then re-mixed with the original soil. The mixed soil was used as one of the treatments a pot experiment (three replicates) as described in Hansen *et al.* (2005). Spring barley (*Hordeum vulgare* L., cv. Evergreen) was sown in April as the test crop and harvested at maturity in August. The grain and straw were each analysed for dry matter content and total N concentration. To estimate the fertilizer replacement value, a reference soil was collected from a neighbouring field with a history of cereal production and placed in a series of pots that received increasing N rates. Initial inorganic N concentration and potentially mineralizable N (anaerobic incubation) were analysed according to Hansen *et al.* (2005).

Data were analysed by one-way analysis of variance (ANOVA) using R (Version 3.1.1). Multiple comparisons were tested with Tukey's *post hoc* method.

## Results

The initial inorganic N concentration of the grassland soils did not differ under the various swards (Figure 1a) but was generally less than in the reference soil, although ammonium:nitrate ratios were relatively greater. In contrast,

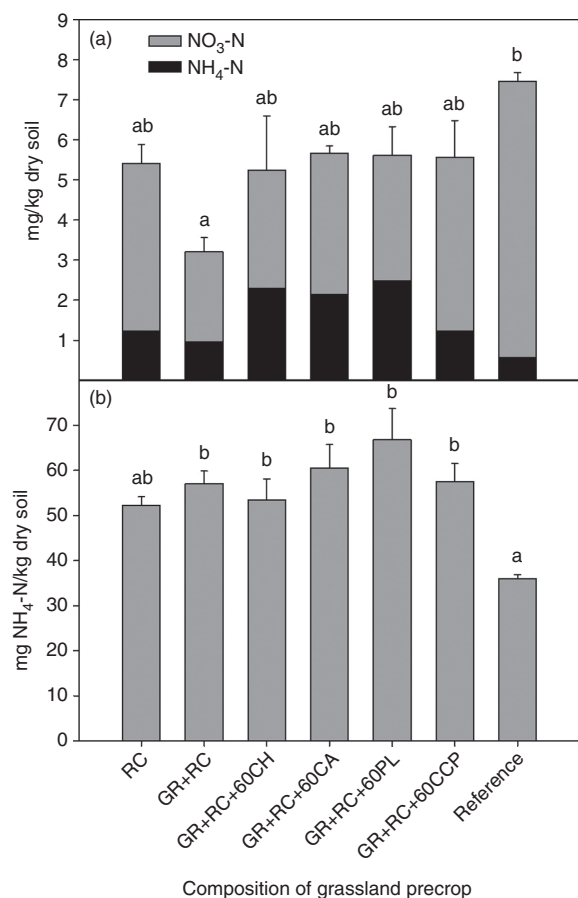
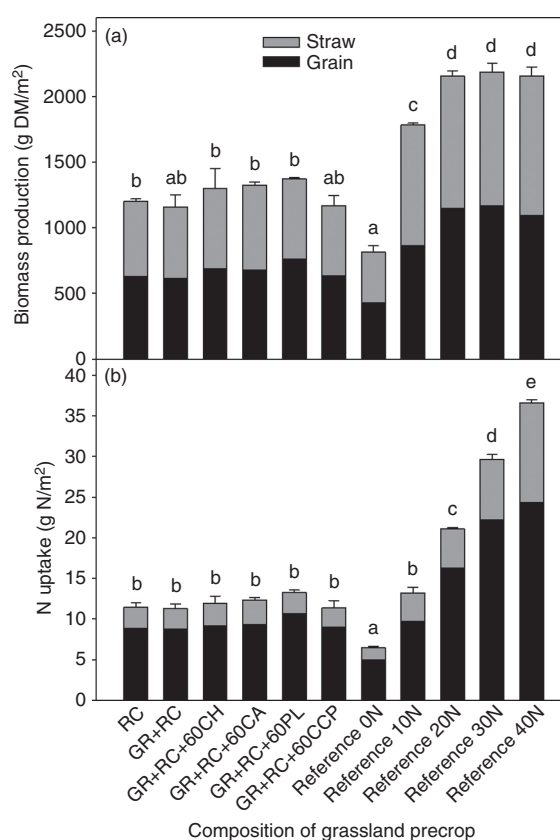
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**Table 1** Species composition (proportion of the seed mixture), and seasonal red clover and total N yield, red clover percentage of total dry matter (DM) production, total N input from N<sub>2</sub> fixation and N balance during the grassland phase in 2014

Seed mixtures	2013					2014				
	Percentage of seed in the mixture sown in 2013					N yield (kg N/ha)		Red clover percent of total DM production	Total N <sub>2</sub> fixation input <sup>1</sup> kg N/ha	N balance <sup>2</sup> kg N/ha
	GR	RC	CH	CA	PL	Red clover	Total			
Pure stand	Red clover		100			480 ± 7 <sup>c</sup>	480 ± 7	100 ± 0 <sup>c</sup>	617 ± 18 <sup>b</sup>	137 ± 19 <sup>c</sup>
Two species	GR+RC	50	50			412 ± 14 <sup>bc</sup>	468 ± 16	82 ± 1 <sup>bc</sup>	594 ± 27 <sup>b</sup>	126 ± 11 <sup>c</sup>
Three species	GR+RC+CH	20	20	60		164 ± 33 <sup>a</sup>	283 ± 34	45 ± 6 <sup>a</sup>	246 ± 50 <sup>a</sup>	-37 ± 16 <sup>ab</sup>
	GR+RC+CA	20	20		60	298 ± 57 <sup>abc</sup>	389 ± 53	66 ± 7 <sup>ab</sup>	450 ± 85 <sup>ab</sup>	60 ± 32 <sup>bc</sup>
	GR+RC+PL	20	20		60	253 ± 49 <sup>ab</sup>	392 ± 52	52 ± 5 <sup>a</sup>	380 ± 72 <sup>ab</sup>	-13 ± 21 <sup>ab</sup>
Five species	GR+RC+60CCP	20	20	20	20	183 ± 53 <sup>a</sup>	324 ± 54	43 ± 7 <sup>a</sup>	270 ± 79 <sup>a</sup>	-54 ± 26 <sup>a</sup>

GR, Perennial ryegrass; RC, Red clover; CH, Chicory; PL, Ribwort plantain; CA, Caraway; CCP, Chicory-Caraway-Ribwort plantain. Values are means (±SE; *n* = 3). Different letters within each column indicate statistically significant (*P* < 0.05) differences between the mixtures. The relative amount of seeds in mixtures were based on the seeding rate of each species in a pure stand. <sup>1</sup>The sum of N fixed in red clover shoot, roots and stubble including fixed N transferred to companion nonlegume species and N immobilized in soil organic N pool calculated using an empirical model (Høgh-Jensen *et al.*, 2004). <sup>2</sup>Difference between N input and removal by harvested aboveground biomass.

**Figure 1** Initial soil inorganic N (a) and net N mineralized in anaerobic incubation (b) in grassland soil (means ± SE; *n* = 3). Different letters indicate significant differences at *P* = 0.05. GR, Perennial ryegrass; RC, Red clover; CH, Chicory; CA, Caraway; PL, Ribwort plantain; CCP, Chicory-Caraway-Ribwort plantain.**Figure 2** Biomass production (a) and N uptake (b) in spring barely test crop grown in the soil previously under different grasslands (means ± SE; *n* = 3). Different letters indicate significant differences at *P* = 0.05. GR, Perennial ryegrass; RC, Red clover; CH, Chicory; CA, Caraway; PL, Ribwort plantain; CCP, Chicory-Caraway-Ribwort plantain. 0N, 10N, 20N, 30N and 40N indicate N fertilizer application, equivalent to 0, 10, 20, 30 and 40 g N/m<sup>2</sup>.

the potentially mineralizable N was significantly larger in the grassland compared with the reference soil ( $P < 0.01$ ) (Figure 1b). However, there were no significant differences among the swards in the potentially mineralizable soil N.

The grain and straw yields of spring barley were similar in the soils from under the different swards (Figure 2a), as were values for N uptake, which ranged from 11 to 13 g N/m<sup>2</sup> (Figure 2b). The total N uptake from the grassland soil under the different swards corresponded to that from the reference soil that received 10 g N/m<sup>2</sup>.

## Discussion

The different species composition of the forb-containing grasslands resulted in large variation in red clover proportion, N<sub>2</sub>-fixation and N yield (Table 1), which previously have been shown to affect the residual N effect (Vrignon-Brenas *et al.*, 2016). We did not observe such an effect in the present pot experiment indicating that inclusion of forbs did not affect the shorter-term N fertility of the grassland precrops. An explanation may be that the N uptake period of spring barley was too short (mainly May and June) to detect differences between mixtures. In the long term this may be different, but it is also possible that including nonlegume forbs in mixtures with competitive forage legumes like red clover will only have a marginal effect on the residual N under mixed-grassland-arable systems with generally a high level of soil fertility.

In conclusion, we showed that inclusion of forbs in perennial ryegrass-red clover mixtures did not affect the short-term residual N available to the subsequent spring barley test crop.

## Acknowledgement

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